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Binding of a monoclonal antibody to B virus gD linear epitope on a whole virus antigen is revealed only after antigen denaturation.

As part of a study for characterization of anti B virus monoclonal antibodies (mAbs), we have discovered a mAb to a B virus glycoprotein D (gD) linear epitope that bound to recombinant gD by Enzyme-linked immunoassay (ELISA), revealed a 50 kDa band by western blot analysis (WBA) but did not bind to the whole virus antigen in ELISA. This unusual reactivity was not seen with other mAbs with anti linear B virus epitopes reactivity.

Based on these results we set up experiments in this study to support our hypothesis that denaturation of BV antigen is required for anti-gD mAb recognition of its epitope. The whole B virus antigens were denatured by different concentrations of SDS and boiling and tested by ELISA and by dot blot analysis (DBA). It was found that the mAb that the whole B virus antigens reacted with the mAb in the DBA after denaturation with 0.005% SDS and boiling, indicating that indeed this epitope is accessible to the mAb only after denaturation. However, this same treatment did not render this epitope detectable by ELISA. We attribute the negative results by ELISA to the lower antigen binding capacity of the polystyrene surface of the ELISA microplate wells as compared to the nitrocellulose antigen binding capacity that is estimated to be 8-10 fold higher.

Key words: B virus, monoclonal antibody, denaturation, ELISA, Dot blot assay, recombinant gD